HEART

Editorial

Dilated cardiomyopathy: a genetic approach

Dilated cardiomyopathy is a myocardial disease characterised by impaired systolic function and dilatation of the left or both ventricles. The disease is not rare, affecting about one in 2500 individuals[1] and, despite the development of new treatments, it remains an important cause of mortality and morbidity and is a leading indication for heart transplantation. The cause of dilated cardiomyopathy is generally unknown and the identification of the aetiological and pathogenetic mechanisms underlying the disease is regarded a research priority.[2]

An important advance in the search for the aetiology has been the recognition of hereditary transmission in a subset of patients, which indicates that in these cases the origin of the disease must be an altered gene product. The recognition of dilated cardiomyopathy as a genetic disease has clinical implications. Furthermore, the development of molecular genetic techniques has provided the tools for the identification of the gene, or genes, causing the disease.

Clinical genetics

A careful evaluation of the familial history of patients with dilated cardiomyopathy and the examination of relatives with a suspected cardiac disease showed that genetic factors were more frequent than previously recognised.[3] Controlled surveys based on the systematic screening of relatives, irrespective of family history, showed a familial form in 20–25% of patients with dilated cardiomyopathy.[4,5] These data probably underestimate the real prevalence, because affected individuals are likely to be missed, particularly in small pedigrees, and because of the absence of early markers of disease and reduced penetrance (the proportion of carriers who manifest the disease). Penetrance is reduced and age-related in dilated cardiomyopathy: as a consequence young family members in particular can appear clinically normal though they carry the disease gene.

Different patterns of transmission and variable clinical features suggest that familial dilated cardiomyopathy (FDC) is the final common pathway of a heterogeneous group of disorders. These may include an autosomal dominant and an autosomal recessive FDC; conduction defects with later development of dilated cardiomyopathy; dilated cardiomyopathies associated with subclinical myopathy, such as the X-linked form; and possibly a mitochondrial dilated cardiomyopathy.

Molecular genetics of familial dilated cardiomyopathies

Molecular genetics offers several approaches to study inherited diseases: *functional cloning*, which allows the identification of gene mutations when the protein defect is known; *positional cloning* (or reverse genetics), based on linkage analysis which allows the chromosomal localisation of the disease genes to be mapped; and the *positional candidate* approach, based on the co-segregation of candidate genes (genes that are candidates for causing the disease) with the disease within families. These approaches have led to impressive advances in the study of primary myocardial diseases during the past few years.

The most frequent form of inherited dilated cardiomyopathy is autosomal dominant FDC which is characterised by development of ventricular dilatation and dysfunction, usually in the second to third decade of life, with progressive heart failure and ventricular arrhythmias. Segregation analysis suggests a monogenic disorder.[4]

The premature mortality, the reduced penetrance of the disease, and the absence of early clinical markers have always been serious obstacles in collecting large enough families for molecular genetic studies. Only recently, two research groups reported the results of linkage analysis performed in families with autosomal dominant FDC. First, a single large kindred was studied by our group: affected family members typically had a poorly contracting left ventricle, often associated with ventricular arrhythmias. Strict diagnostic criteria, based on full invasive evaluation of the affected family members, were used to assign clinical status.[2]

After many of the candidate genes were excluded, a whole-genome random screening with more than 250 polymorphic markers was undertaken. This allowed the analysis of about 95% of the genome. Linkage of autosomal dominant FDC was eventually found with chromosome 9 (9q13–q22) in this kindred and in two other families[6] (fig 1). A second locus for autosomal dominant FDC was then localised on chromosome 1q32 in a large Utah family[7] (fig 1). The analysis of the candidate genes mapping in these regions is currently under investigation.

A rare form of FDC is characterised by autosomal dominant cardiac conduction system disease and later development of myocardial dysfunction (called conduction disease and dilated cardiomyopathy, CDDC). The affected family members manifest arrhythmias and atrioventricular block in the second to third decade of life and a progressive cardiomegaly and heart failure in the fifth to sixth decade. A first linkage study carried out in a single large family from Ohio mapped the CDDC disease gene in the centromere of chromosome 1 (1p1–1q1)[8] (fig 1). Recently, in another family of Swiss–German ancestry and similar clinical features, linkage was found with the short arm of chromosome 3 (3p22–p25)[9] (fig 1). The disease genes are unknown and are under investigation.

Till now the only inherited dilated cardiomyopathy for which the disease gene was known is X-linked dilated cardiomyopathy (XLDC), which generally presents in teenage boys as rapidly progressive congestive heart failure
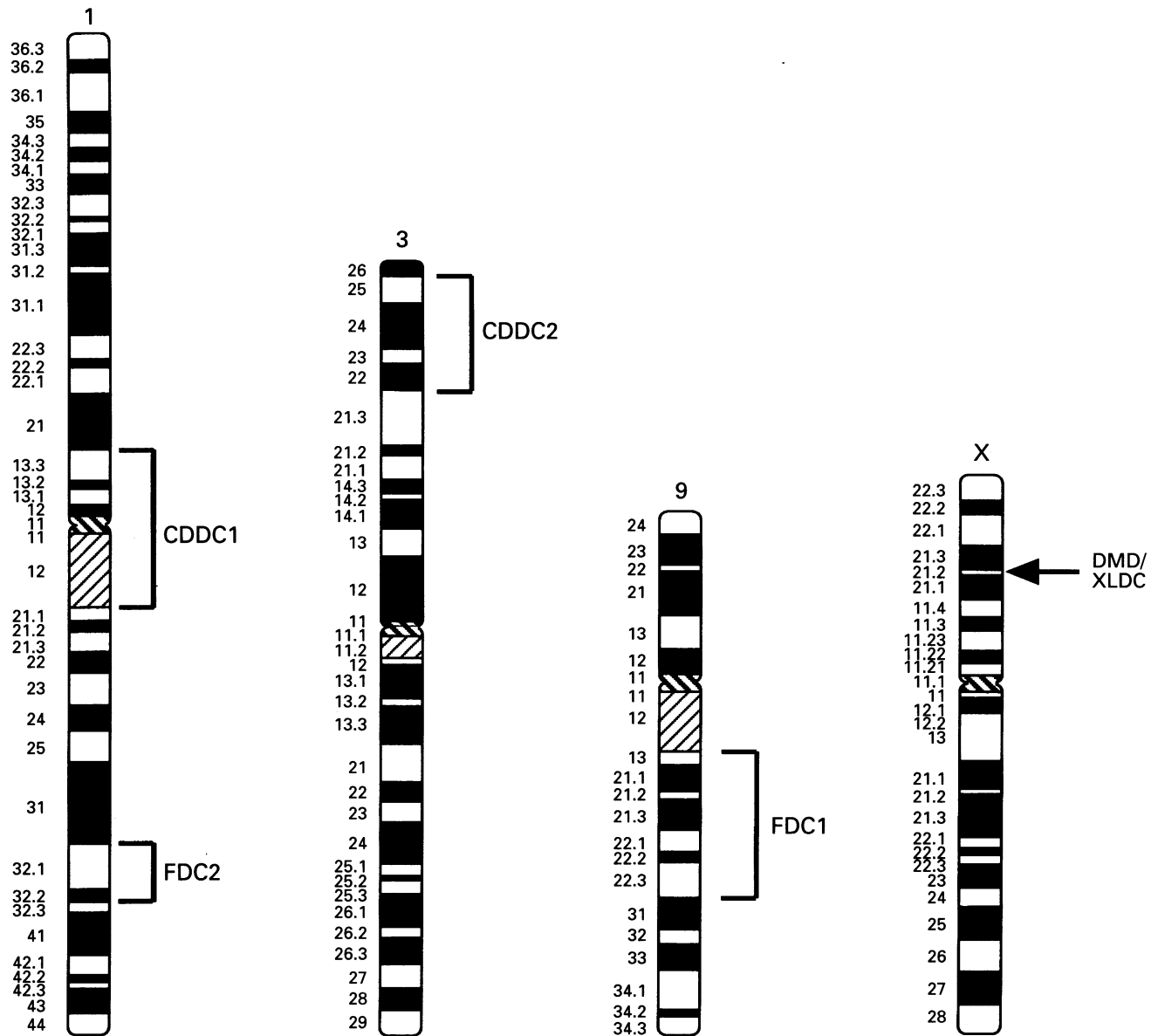
Figure 1 Diagrams of the four chromosomes containing known disease gene loci for different forms of dilated cardiomyopathy. The disease loci, identified by square brackets, were mapped on chromosome 1, containing the CDDC1 locus (1q1–p1)[8] and the FDC2 locus (1q31-q32)[7], chromosome 3 with the CDDC2 locus (3p22-p25)[9], chromosome 9 with the FDC1 locus (9q13-q22)[6], and chromosome X with the dystrophin gene (DMD), cause of XLDC (Xp21) (reprinted from Mestroni et al,[17] with the permission of Archives des Maladies du Coeur et Vaisseaux).

in the absence of clinical signs of skeletal myopathy. Affected family members may have a mild increase of muscle creatine kinase (MM-CK). Female carriers have a later onset and slower progression of the disease. This condition is characterised by the transmission of the disorder with the X chromosome (no male-to-male transmission) in a dominant or recessive fashion.

Two independent studies showed that the dystrophin gene, the same one that causes Duchenne and Becker muscular dystrophies, was also responsible for XLDC[10 11] (fig 1). The identification of deletions in the region of the muscle promoter–first muscle exon[11] and recently of a point mutation (G/T) in the 5' splice site of the first intron[12] (fig 2) leads to the hypothesis that the integrity of the 5' region of the gene is critical for the expression of dystrophin in the heart. Immunocytochemical studies fully explain the phenotype of these patients, showing that dystrophin is reduced in quantity but normally distributed in skeletal muscle, whereas it is undetectable in the cardiac muscle.[12] Studies of the expression of the major dystrophin mRNA isoforms show none in the myocardium,

whereas brain and Purkinje cell (but not muscle) isoforms are detectable in the skeletal muscle.[12]

Furthermore, deletions of exons 48 and 49 were found to be frequently associated with cardiomyopathy in patients with Becker muscular dystrophy.[13] Our data indicate that carriers of these deletions can present as isolated dilated cardiomyopathy in absence of overt signs of myopathy (unpublished data).

By analogy, other cytoskeletal proteins could be involved in the aetiology of dilated cardiomyopathy. Deficiency of adhalin, a dystrophin-associated glycoprotein, has recently been described in a patient with dilated cardiomyopathy associated with signs of muscle dystrophy.[14] Furthermore, alterations of the vinculin gene, coding for a protein of the intercalated disk, have been reported in a patient with dilated cardiomyopathy.[15]

Dilated cardiomyopathy is found in mitochondrial diseases, usually complex syndromes caused by mitochondrial DNA (mtDNA) defects and inherited by maternal transmission. Multiple deletions of mtDNA have been described in a single family with dilated cardiomyopathy,[16] as well as in
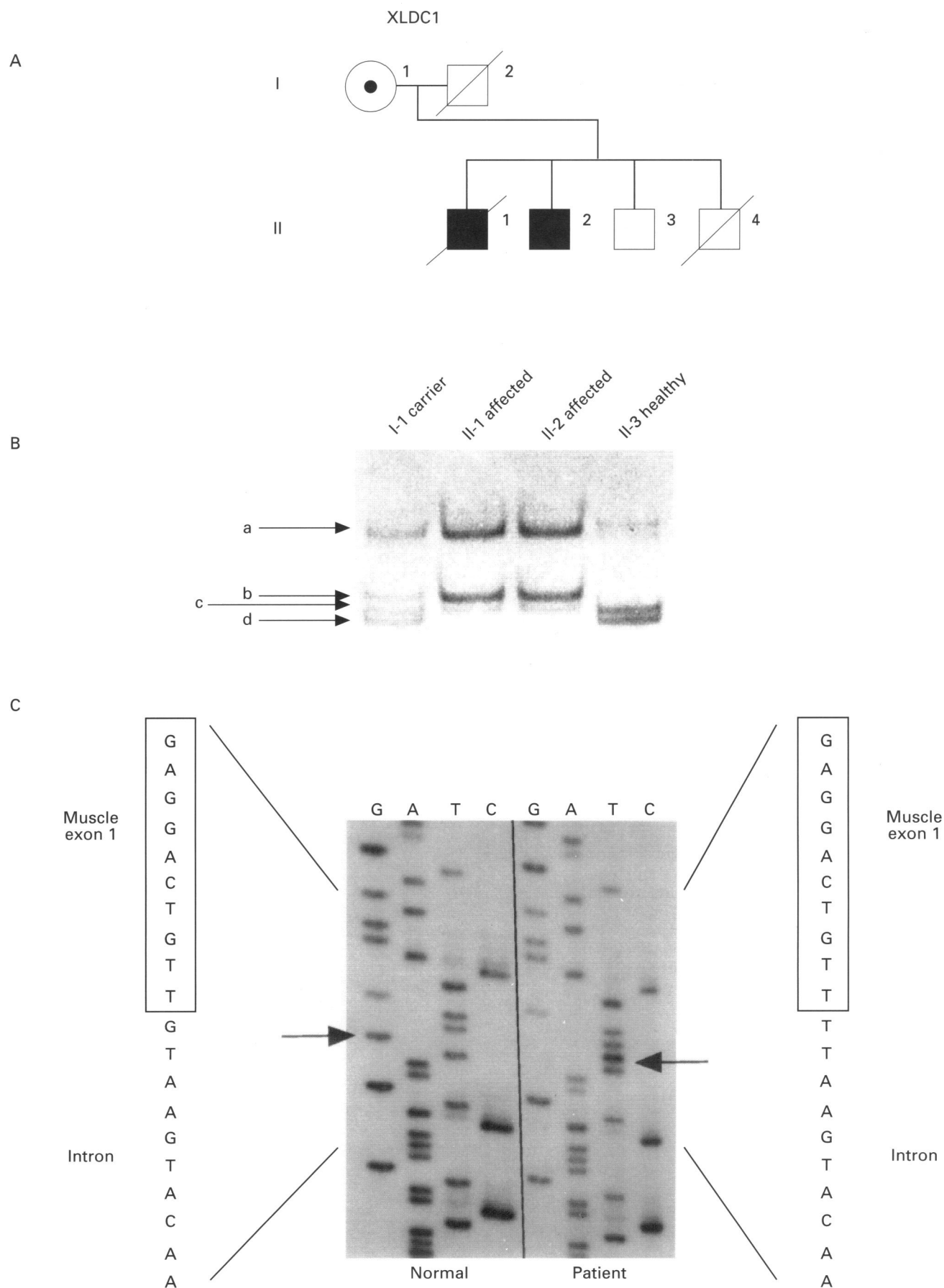
Figure 2 XLDC caused by a dystrophin gene point mutation. (A) Pedigree of family XLDC1. Individuals are indicated by generation and pedigree number. Their status is indicated by solid symbols (affected), open symbols (unaffected), and a circle with a dot (carrier). (B) Mutation analysis: SSCP analysis of the PCR product encompassing the first muscle exon-intron junction. Individual ssDNA strands are indicated by arrows (a to d). Individuals II-1 and II-2 (affected) carry the same dystrophin allele, which is different from the one of individual II-3 (normal). The mother (I-1) is heterozygous, as expected. (C) Sequence analysis of the first muscle exon-intron junction of dystrophin gene. In the patient II-2 a G/T point mutation eliminates the 5' splice site consensus sequence. The normal control is represented by the healthy brother (II-3) (reprinted from Milasin et al,[12] with the permission of Oxford University Press).

sporadic dilated cardiomyopathy, however, their primary role as pathogenetic factors is still controversial.

Clinical implications of the genetic and molecular studies

The fact that several surveys show that dilated cardiomyopathy is a genetic disease in a significant proportion of patients and that a gene defect should be considered as the cause of the disease in at least one third of patients has important clinical implications.

The management of a potentially inherited disorder requires an accurate investigation of the familial history and a systematic screening of first degree relatives (parents, siblings, and offspring). Family studies allow early clinical diagnosis and for treatment to be started before symptoms develop. Moreover, a molecular diagnosis of XLDC and of the carrier status is now possible. In the future, linkage analysis could be used as a diagnostic procedure for the detection of family members at risk of the disease. Moreover, genetic counselling should be offered to patients with dilated cardiomyopathy to inform them about the characteristics of the disease, the risk in their relatives, the scope of early treatment, and the likelihood of future developments.

On the other hand, the experience in family studies raises new questions about the diagnostic criteria. Frequently, relatives show minor signs of myocardial disease, such as segmental wall motion abnormalities, dilatation without ventricular dysfunction, frequent and repetitive arrhythmias, and conduction defects that do not satisfy the traditionally accepted diagnostic criteria of dilated cardiomyopathy. These signs in the context of a hereditary disease strongly suggest early manifestations of the disorder. Follow up studies and, eventually, a molecular diagnosis will resolve these questions. The possibility of subclinical skeletal muscle involvement should always be considered, and in suspected cases supplementary diagnostic procedures, such as skeletal muscle immunocytochemistry and quantitative electromyography, should be performed.

The identification of the genes and the understanding of the molecular mechanisms causing dilated cardiomyopathy will provide a means not only of diagnosing and preventing the disease but also of developing genotype-based treatments to modify the primary defect underlying the disease.

Recently, another locus for autosomal dominant FDC has been reported on the short arm of chromosome 10 (10q21–23).[18] This form of FDC appears to be characterised by the association with mitral valve prolapse and by a high penetrance.

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